AD NUMBER
AD302603
CLASSIFICATION CHANGES
TO: unclassified
FROM: secret
LIMITATION CHANGES
TO: Approved for public release, distribution unlimited
FROM: Distribution authorized to DoD and DoD contractors only; Foreign Government Information; 16 SEP 1958. Other requests shall be referred to British Embassy, 3100 Massachusetts Ave., NW, Washington, DC 20008.
AUTHORITY
WO 195/14431, 8 Oct 2009; WO 195/14431, 8 Oct 2009

DISCLAIMER NOTICE

THIS DOCUMENT IS BEST QUALITY PRACTICABLE. THE COPY FURNISHED TO DTIC CONTAINED A SIGNIFICANT NUMBER OF PAGES WHICH DO NOT REPRODUCE LEGIBLY.

SECRET

rmed Services Technical Information Agency

ARLINGTON HALL STATION
ARLINGTON 12 VIRGINIA

OR
CRO-CARD
ONTROL ONLY

1 OF 1

SECRET

302603

SECRET DISCREET

P.T.P. 443

Copy No. 80

MINISTRY OF SUPPLY

581423

DIRECTORATE OF CHEMICAL DEFENCE RESEARCH AND DEVELOPMENT

CHEMICAL DEFENCE EXPERIMENTAL ESTABLISHMENT

THE EFFECTS OF GB AND VX ON THE ISOLATED RABBIT HEART AND THEIR REVERSAL BY P2S AND ATROPINE

BERYL M. ASKEW

PORTON TECHNICAL PAPER No. 64

C.D.E.E.
Porton.
Wilts.

SECRET DISCREET

SECRET DISCREET

PORTON TECHNICAL PAPER No.643:
COPY No. 80
DATE: 16 SEP 1958

THE EFFECTS OF GB AND VX ON THE ISOLATED RABBIT HEART AND THEIR REVERSAL BY P2S AND ATROPINE

By

Beryl M. Askew

SUMMARY

1. Perfusion of the isolated rabbit heart with GB or VX at concentrations of 0.5 - 0.005/ug/ml caused a reduction in heart rate to a mean of approximately 70% of normal. There was virtually no change in the amplitude of contraction.

2. When GB or VX were perfused together with 10 m/ug/ml ACh the reduction in heart rate was more marked, whilst in a few cases the heart ceased to beat. There was, in addition, a decrease in the amplitude of contraction.

3. P2S (25/ug/ml) caused a return to 80-90% of the initial heart rate within 30 minutes, both when the agents had been used alone or together with ACh. With atropine (2/ug/ml) recovery of heart rate was considerably faster and reached a mean of at least 90% within 2-3 minutes.

4. P2S or atropine also reversed the amplitude changes produced by GB or VX together with ACh.

5. There was no evidence to suggest that either GB or VX caused any myocardial damage at the concentrations used.

(Sgd.) C. Lovatt Evans,
Head, Physiology Section.

BMA/MB

(Sgd.) W.S.S. Ladell,
Supt., Medical Division.

SECRET DISCREET

SECRET

PORTON TECHNICAL PAPER No.643

COPY No. 80

DATE: 16 Sep 1958

THE EFFECTS OF GB AND VX ON THE ISOLATED RABBIT HEART AND THEIR REVERSAL BY P2S AND ATROPINE

By

Beryl M. Askew

INTRODUCTION

It has frequently been shown that the organophosphorus compounds cause both a fall in blood pressure and reduction in heart rate in most species (1, 2). However, few direct studies have been carried out on the effect of such compounds on the isolated perfused heart. Quilliam and Strong (3) using DFP, and Salerno and Coon (4) using DFP, HETP and TEPP found some decrease in the amplitude of contraction of the heart, but little or no effect on heart rate even when relatively high concentrations were used. However, the depressant action of ACh on the heart was markedly potentiated. The changes in amplitude found were not altered by prior treatment of the heart with atropine. Similarly it has been reported (5) that GB, whilst causing bradycardia in the dog heart-lung preparation, was without effect even in large doses, on the isolated rabbit heart. Briscoe and Burn (6) found that a number of antiChE agents including DFP were effective in reducing the rate of spontaneously beating rabbit auricles and recently Larson and Brown (7) showed that both heart rate and amplitude of contraction of the isolated rabbit heart were sharply decreased by V-agents.

In the work reported here a comparison has been made of the effects of isopropyl methylphosphonofluoridate (GB) and S-2-diisopropylaminoethyl-O-ethyl methylphosphonothiolate (VX) on the isolated rabbit heart. Since free ACh is found in the blood of rabbits after single doses of antiChE compounds (Barnes and Duff (8), Stewart (9)) at a level of approximately 10 mμg/ml, the effect of including this concentration of ACh in the perfusion fluid, on the response of the heart to GB or VX was also studied. The ability of 2-hydroxyiminomethyl-N-methylpyridinium methanesulphonate (P2S) and atropine to reverse these effects has been investigated.

MATERIALS AND METHODS

Female rabbits of weight 1.5-2.5 kg were killed by a blow on the head and the heart was rapidly removed, washed and set up for perfusion through the aorta by the method of Langendorff. The perfusion pressure for each experiment was kept constant by the use of Marriot bottles, pressures of

38-45 cm water being used. The hearts were perfused at 37°C by McEwen's solution*(10) gassed with 95% O_2 - 5% CO_2 to give a pH of 7.4-7.5. The heart was partially enclosed in a water jacket kept at 37°C to keep local fluctuations in air temperature to a minimum.

A thread connected the apex of the heart via 2 pulleys to the recording lever. Electrical contacts from this lever activated a digital counter which was used to count the heart rate over 30 second periods. Flow through the coronary vessels was measured using a drop recorder and Thorpe impulse counter. The ECG was recorded on an Ediswan Pen Recorder from two wick electrodes applied to the right and left ventricles of the heart.

The hearts were perfused for approximately 30 minutes with McEwen's solution alone by which time the rate and amplitude of contraction had become virtually constant. The agent, GB or VX, dissolved in McEwen's solution was then perfused from a second reservoir for a period of 30 minutes, followed by McEwen's solution either alone or containing P2S (25 µg/ml) or atropine sulphate (2 µg/ml), for a further 30 minutes. In experiments in which ADh was used, the concentration in the perfusion fluid was 10 m µg/ml.

The heart rate at the commencement of each experiment was taken as 100% and all subsequent rates were expressed as a % of the initial rate. Changes in the amplitude of contraction were expressed in the same manner. In a few cases rabbits were pretreated with reserpine. They received 1.5 mg/kg i.p. 48 hours before use and a further 5 mg/kg i.v. 24 hours later, as suggested by Burnand Rand (11).

RESULTS

1. Control Experiments

After the stabilization period of approximately 30 minutes the mean heart rate was 84/30 sec (S.D. = ± 14; 136 hearts) 5 control experiments were carried out to determine what changes in heart rate and amplitude of contraction occurred over a further period of 75 minutes. For the first 30 minutes of this period the heart was perfused with McEwen's solution alone; the perfusion fluid was then changed to one containing P2S (25 µg/ml) or atropine (2 µg/ml). This enabled comparisons to be made between (a) normal heart/agent treated heart and (b) normal heart treated with P2S or atropine/agent treated heart plus P2S or atropine. During this 75 minute period the heart rate fell progressively to a mean of 92% (range 85-95%), the rate of decrease being unaffected by the addition of P2S or atropine.

The amplitude of contraction increased to a mean of 109% by the end of the 30 minute perfusion with McEwen's solution alone. On changing to perfusion from a second reservoir, a temporary increase or decrease in amplitude always occurred regardless of the content of the perfusion fluid. Attempts to eliminate this artefact by various means were unsuccessful, but the amplitude returned to its previous level within 10-15 minutes and thereafter remained virtually constant in the control

*NaCl 7.6 g, KCl 0.42 g, $CaCl_2$ 0.24 g, NaH_2PO_4 0.143 g, $NaHCO_3$ 2.1 g, dextrose 2.0 g, sucrose 4.5 g, distilled water to 1,000 ml.

experiments. At the end of the 75 minute period (which included the period with P2S or atropine) the mean amplitude was 106% (range 100 - 114%). ACh at the concentration used of 10 m/ug/ml was found to be without effect on the heart rate although it caused a very slight gradual increase in the amplitude of contraction.

2. Effect of GB and VX on heart rate and amplitude of contraction

Hearts were perfused for periods of 30 minutes with GB or VX at concentrations of 0.5, 0.05 and 0.005 /ug/ml respectively. In all cases the agents were found to produce varying degrees of bradycardia. Fig.1 shows the mean effect of the two agents at these three concentrations; detailed figures from which these curves were drawn are given in Table AI, Appendix I. GB, 0.005/ug/ml had a less marked effect on rate than either of the two stronger concentrations. With VX the effect on rate after 30 minutes perfusion was the same for all three concentrations. However, with both agents at each dose level there was a marked variation in the response of different hearts which was independent of the heart rate at the commencement of the experiment. For example, with 0.5/ug/ml GB (22 hearts) the rate after 30 minutes had fallen to a mean of 67.3% with a range of 43.5 - 83.5%. For the same concentration of VX the mean rate for 15 hearts was 72.5% with a range of 61.0 - 83.0%.

As shown in Fig.2, GB and VX had little effect on the amplitude of contraction even at the strongest concentration used (0.5/ug/ml).

3. Effect of GB and VX in presence of ACh on heart rate and amplitude

In the presence of ACh, 10 m/ug/ml, GB and VX at concentrations of 0.05 and 0.005/ug/ml produced a marked bradycardia as shown in Fig.1 In a few cases there was a complete cessation of heart beat within the 30 minute period. With GB, 2 out of 11 and 1 out of 13 hearts stopped with concentrations of 0.05 and 0.005 ug/ml respectively. Similarly 1 out of 12, and 1 out of 7 hearts stopped with the corresponding concentrations of VX. (Only hearts which continued to beat throughout the full 30 minutes period are included in the graphs).

In the presence of ACh, there was a progressive decrease in the amplitude of contration of the heart to a mean of 67% with GB (0.05/ug/ml) and 73% with VX (0.05/ug/ml) at the end of 30 minutes (Fig.2).

4. Effect of GB and VX in presence of ACh on the ECG

In view of the marked bradycardia which occurred when GB or VX were perfused together with ACh, ECG recordings were taken from some hearts to determine whether there was any gross change in the ECG. In control experiments slight changes in the amplitude of the different components of the ECG occurred over a period of time although the basic pattern remained unaltered. In the majority of cases the only further change seen in the ECG after perfusion with GB or VX together with ACh was an increase in the S-T interval concomitant with cardiac slowing (Fig.3,(b)). In a very few cases the P wave disappeared whilst the QRS complex became altered in shape. It was, however, followed by a modified T wave (Fig.4 (c)) after the normal time interval suggesting that the A-V node had taken over the function of pacemaker.

5. The ability of P2S and atropine to reverse the effects of GB or VX alone and with ACh.

When hearts were perfused with McEwen's solution alone after a period of perfusion with GB or VX, there was no recovery in heart rate. However, as shown in Fig.5 and in detail in Table AII, Appendix I, P2S (25 µg/ml) caused a recovery of heart rate to 80-90% of the initial rate within 30 minutes regardless of whether the GB or VX had been used alone or with ACh. (Whenever ACh was used with GB or VX, the same concentration of ACh was included in the P2S and atropine solutions).

With atropine (2 µg/ml) the rate of recovery was considerably faster and had reached a maximum within 2-3 minutes; as shown in Table I the degree of recovery was also slightly greater since the heart rate returned, in the absence of ACh, to a mean of at least 95% of normal.

Table 1

Effect of Atropine on the Bradycardia produced by GB or VX

GB or VX Perfused from 0'-30'

Atropine (2 µg/ml) perfused from 30' onwards

ACh 10 m µg/ml Fig. in brackets = S.D.

Agent	Conc. (µg/ml)	Number of hearts	Mean heart rate (% of rate at 0' min)		
			30'	35'	45'
GB	0.5	7	70.1(± 8.5)	105.6(± 8.9)	104.7(± 9.6)
	0.005	4	83.6(± 3.2)	97.1(± 2.4)	95.5(± 2.4)
GB + ACh	0.05	5	31.0(± 14.5)	90.6(± 6.3)	88.5(± 5.4)
	0.005	3	55.0(± 18.0)	96.8(± 5.3)	94.7(± 5.7)
VX	0.5	4	74.8(± 6.3)	96.5(± 5.2)	98.0(± 6.0)
	0.005	4	66.9(± 10.7)	95.3(± 3.3)	95.3(± 2.8)
VX + ACh	0.05	2	48.0	89.0	88.3

The decrease in amplitude which occurred when the agents were perfused together with ACh was completely reversed by both P2S and atropine as shown in Table II.

Table II

Reversal of Amplitude changes by P2S and Atropine

GB / VX + ACh (10 µg/ml) } perfused from 0' - 30'

Atropine (2 µg/ml) or P2S. (25 µg/ml) } perfused from 30' onwards

	Agent (0.05 µg/ml)	Mean amplitude of contraction (% of amp. at 0')					
		30'	40'	45'	50'	55'	60'
P2S	GB	79.4	93.8	95.4	95.4	98.6	99.8
	VX	75.9	97.7	100.4	103.0	103.4	102.9
Atropine	GB	68.5	95.0	95.0	96.2	97.8	98.5
	VX	79.8	102.8	104.5	104.5	104.0	104.0

Where marked ECG changes had occurred after perfusion with GB or VX and ACh, atropine and P2S restored the ECG to normal (Figs. 3 and 4).

6. Coronary flow

GB and VX at the concentrations used were without effect on the rate of coronary flow.

7. The effect of pretreatment of rabbits with reserpine on the bradycardia produced by GB

The effect of 0.5 µg/ml GB on hearts of rabbits pretreated with reserpine to deplete the heart of its stores of adrenaline and nor-adrenaline (12) was investigated in order to determine whether the marked variation in the degree of bradycardia produced was due to a release of adrenaline. The results are given in Table III.

Table III

Effect of pretreatment of rabbits with reserpine on bradycardia produced by 0.5 µg/ml GB

	Number of hearts	Mean heart rate (% of initial rate) and S.D.						
		5'	10'	15'	20'	25'	30'	Range
No reserpine	22	79.2 (± 10.4)	70.5 (± 11.7)	68.7 (± 11.0)	68.3 (± 10.5)	67.6 (± 10.5)	67.3 (± 10.8)	43.5-83.5
Reserpine	10	85.5 (± 12.8)	69.5 (± 14.4)	65.3 (± 12.7)	63.4 (± 13.0)	63.2 (± 12.5)	62.4 (± 11.3)	50.0-75.5

As will be seen from the table, there was no significant difference between the two groups of hearts.

DISCUSSION

Perfusion of the isolated rabbit heart with GB or VX at concentrations of 0.5 - 0.005 µg/ml caused a reduction in heart rate but little change in the amplitude of contraction. When the agents were perfused together with 10 mµg/ml ACh (a concentration which had no deleterious effect on control hearts) the reduction in rate was more marked and there was in addition a decrease in the amplitude of contraction. The susceptibility of different hearts to the action of the same concentration of GB or VX, however, showed a fairly wide variation. In similar experiments with eserine, Brisco and Burn(13) also commented on the variability between different hearts.

Results obtained by Hoffmann, Hoffmann, Middleton and Talesnik (14) on the isolated heart, led these authors to suggest that ACh acts on certain intracardiac structures and stimulates them to release adrenaline. Since adrenaline will produce an increase in heart rate and amplitude of contraction, it seemed possible that the variation in response of individual hearts to the action of GB and VX might be due to a variable release of adrenaline by these agents. The effect of 0.5 µg/ml GB on hearts from rabbits pre-treated with reserpine to deplete their stores of adrenaline and noradrenaline was therefore investigated in order to determine whether there would then be a more constant response. However, since the S.D. of 10 hearts from reserpine-treated animals was as great as that for normal hearts (Table III) it seems unlikely that the release of adrenaline from the heart could account for the variation found.

Bulbring and Burn(15) working with isolated auricles obtained results which suggested that the normal rhythmic contractions were dependent on the synthesis of ACh. Burn and Kottegoda (16) later investigated the action of eserine on isolated auricles, since if ACh controls the rhythmic movements of the heart, then the ChE present might be expected to play a part in controlling activity by destroying the ACh. They found that eserine caused a reduction in the rate of spontaneously beating auricles, suggesting that there is an optimal concentration of ACh for the maintenance of the beat and that amounts in excess of this depress the rate.

It has been shown by Hutter and Trautwein (17) that in the sinus venosus of the frog heart during diastole a slow depolarisation, the pacemaker potential, develops. Marshall and Vaughan Williams (18) similarly found that in the rabbit auricle the pacemaker produces regularly occurring potentials to which the rest of the auricle can respond with propagated action potentials. It is suggested (Burn and Rand (19)) that the synthesis of ACh by choline acetylase maintains the resting membrane potential of the auricle at a threshold level for a propagated response. This resting potential is increased by the application of ACh or carbamylcholine (Burgen and Terroux (20)). AntiChE compounds might also be expected to cause an increase in the resting potential by causing an accumulation of ACh. If this is so, such compounds would be expected to slow the heart since the pacemaker will have to undergo a greater degree of depolarisation in order to reach the critical level necessary for the production of a propagated action potential.

With the exception of the most dilute concentration of GB, all concentrations of GB and VX were found to reduce the heart rate to a mean of approximately 70% of the initial rate at the end of the 30-minute perfusion period. As calculated from the 2nd order rate constants for both inhibitors at 37°C, pH 7.4 - 7.6, the strongest concentration used i.e. 0.5 µg/ml, is about 100 times greater than the minimum concentration which would be expected to produce total inhibition in vitro of both true and pseudo-ChE within 30 minutes. Assuming that the heart ChE is reasonably accessible to GB and VX, the activity of the isolated heart after 30-minutes perfusion with these agents should therefore represent that occurring in the absence of any ChE activity. When 10 m µg/ml ACh was present in the perfusion fluid in addition to GB or VX, the bradycardia produced was considerably more marked, whilst in one or two cases the heart ceased to beat. This greater effect would be expected since the concentration of ACh around the pacemaker must be higher and in the presence of little or no ChE, the rhythmic activity of the pacemaker will depend on the rate at which ACh can diffuse away from the site of action.

It appears that the heart can continue to beat in the absence of ChE activity, provided that much of the excess ACh can be removed via the coronary circulation. The difference in the degree of slowing produced in different hearts by the same concentration of GB or VX might be explained by a difference in the sensitivity of the auricle in the region of the pacemaker to the action of ACh. Alternatively, the amount of ACh synthesised by the heart, whilst constant for individual hearts, may vary with different hearts. It has been shown for instance by Bulbring, Kottegoda and Shelley (21) that the absolute amount of true and pseudo-ChE activity varied widely in auricles from different rabbits.

When hearts were perfused only with McEwen's solution after a period of perfusion with GB or VX, there was no increase in heart rate. Atropine (2 µg/ml) however, rapidly restored the rate to at least 95% of the initial rate. It has been shown by Burgen and Terroux (20) that following the action of ACh or carbamylcholine on the auricle, atropine will reduce the resting membrane potential approximately to normal. Concomitant with this action of atropine a corresponding increase in heart rate would be expected. Burn and Rand (19) say in their discussion 'The conception that choline acetylase normally forms ACh to maintain the membrane potential has given rise to the question why the contractions of the atria under normal circumstances are not arrested by atropine. The rapid action of atropine on vagal stimulation may be due to vagal ACh being liberated at a point which is extracellular. ACh formed constantly by the atria may be intracellular and therefore not so readily neutralized'.

25 µg/ml P2S is the peak blood level found in rabbits approximately 10 minutes after an intramuscular injection of 50 mg/kg (22). This concentration of P2S produced a recovery in heart rate to a mean of 80-90% of the initial rate within 30 minutes for all concentrations of GB and VX both alone and with ACh. The slower action of P2S compared with atropine in restoring the heart rate towards normal could be explained by the time taken to reactivate sufficient ChE to remove the excess ACh.

GB and VX had virtually no effect on the amplitude of contraction of the heart and there was no indication of myocardial damage. Although in the presence of ACh, GB and VX caused a fairly marked reduction in amplitude this was rapidly reversed both by P2S and atropine. Furthermore, there was no apparent difference between the actions of GB and VX and it has previously been shown by Martin and Wills (23) that GB causes no myocardial damage.

SUMMARY

1. Perfusion of the isolated rabbit heart with GB or VX at concentrations of 0.5 - 0.005 µg/ml caused a reduction in heart rate to a mean of approximately 70% of normal. There was virtually no change in the amplitude of contraction.

2. When GB or VX were perfused together with 10 m/µg/ml ACh the reduction in heart rate was more marked, whilst in a few cases the heart ceased to beat. There was, in addition, a decrease in the amplitude of contraction.

3. P2S (25 µg/ml) caused a return to 80-90% of the initial heart rate within 30 minutes, both when the agents had been used alone or together with ACh. With atropine (2 µg/ml) recovery of heart rate was considerably faster and reached a mean of at least 90% within 2-3 minutes.

4. P.S or atropine also reversed the amplitude changes produced by GB or VX together with ACh.

5. There was no evidence to suggest that either GB or VX caused any myocardial damage at the concentrations used.

ACKNOWLEDGEMENT

Miss J. Stratton gave technical assistance.

(Sgd.) C. Lovatt Evans,
Head, Physiology Section.

BM/MB

(Sgd.) W.S.S. Ladell,
Supt., Medical Division.

Appendix I

Table AI

Effect of GB and VX on heart rate

Fig. in brackets = S.D.

ACh. = 10 m/ug/ml

Agent	Conc (/ug/ml)	No. hearts	Mean heart rate (% of rate at 0 min)					
			5'	10'	15'	20'	25'	30'
GB	0.5	22	79.2 (± 10.4)	70.5 (± 11.7)	68.7 (± 11.0)	68.3 (± 10.5)	67.6 (± 10.5)	67.3 (± 10.8)
	0.05	6	90.4 (± 5.9)	76.4 (± 7.8)	69.5 (± 4.3)	67.3 (± 2.9)	67.2 (± 3.2)	65.6 (± 2.2)
	0.005	13	99.3 (± 2.1)	91.8 (± 5.4)	86.2 (± 7.7)	82.3 (± 9.2)	81.1 (± 9.7)	79.8 (± 10.2)
GB + ACh	0.05	9	81.9 (± 9.2)	54.8 (± 16.9)	43.6 (± 13.2)	37.0 (± 12.9)	36.0 (± 13.0)	35.8 (± 14.2)
	0.005	12	92.5 (± 7.8)	80.6 (± 15.8)	69.3 (± 14.0)	63.4 (± 13.3)	56.8 (± 12.6)	53.4 (± 12.7)
VX	0.5	15	88.4 (± 6.1)	79.1 (± 6.0)	76.3 (± 5.9)	74.2 (± 6.2)	73.1 (± 7.0)	72.5 (± 6.6)
	0.05	8	88.3 (± 6.3)	77.4 (± 7.6)	73.2 (± 6.6)	72.1 (± 6.2)	71.6 (± 5.6)	70.2 (± 5.9)
	0.005	12	95.8 (± 2.9)	86.0 (± 7.1)	77.9 (± 6.8)	74.2 (± 8.4)	72.0 (± 8.3)	70.2 (± 8.8)
VX + ACh	0.05	10	84.5 (± 8.6)	59.7 (± 9.2)	52.4 (± 12.4)	45.5 (± 16.0)	43.7 (± 17.2)	41.7 (± 18.4)
	0.005	6	90.1 (± 2.4)	72.8 (± 11.0)	55.3 (± 18.7)	46.5 (± 18.7)	41.7 (± 19.9)	38.8 (± 21.1)

Appendix I

Table AII

Effect of P2S on bradycardia produced by GB and VX

GB or VX perfused from 0' - 30'.

P2S (25/ug/ml) perfused from 30' onwards.

ACh 10 m/ug/ml.

Fig. in brackets = S.D.

Agent	Conc (/ug/ml)	No. hearts	Mean heart rate (% of rate at 0 min)						
			30'	35'	40'	45'	50'	55'	60'
GB	0.5	7	56.9 (± 8.1)	63.4 (± 7.8)	73.5 (± 7.7)	77.8 (± 7.1)	79.7 (± 5.7)	80.9 (± 6.0)	82.4 (± 4.9)
	0.005	4	72.0 (± 6.7)	83.4 (± 7.7)	84.4 (± 6.6)	83.9 (± 5.6)	85.1 (± 6.0)	84.3 (± 5.2)	83.3 (± 5.4)
GB + ACh	0.05	3	46.5 (± 10.8)	79.0 (± 1.5)	82.0 (± 3.9)	81.3 (± 3.7)	82.0 (± 3.1)	83.0 (± 3.1)	83.7 (± 2.8)
	0.005	5	55.4 (± 14.0)	77.1 (± 9.3)	79.8 (± 7.4)	80.4 (± 7.4)	82.7 (± 7.0)	83.9 (± 6.3)	83.2 (± 6.1)
VX	0.5	9	69.4 (± 5.5)	75.7 (± 5.0)	78.7 (± 5.4)	80.2 (± 5.2)	81.6 (± 5.5)	81.4 (± 5.2)	82.0 (± 5.6)
	0.005	6	78.3 (± 5.2)	81.2 (± 6.3)	84.4 (± 4.3)	86.8 (± 4.6)	87.4 (± 5.1)	87.5 (± 6.1)	87.5 (± 6.1)
VX + ACh	0.05	5	43.6 (± 9.0)	64.3 (± 5.8)	78.2 (± 7.1)	79.3 (± 8.1)	81.8 (± 8.1)	81.4 (± 6.8)	81.9 (± 7.8)
	0.005	4	29.3 (± 19.5)	64.1 (± 21.4)	79.1 (± 5.4)	79.6 (± 6.0)	85.1 (± 4.9)	87.0 (± 4.7)	87.5 (± 5.2)

Appendix II

The effects of V agents on isolated perfused rabbit hearts.

Comments on CWLR 2148 and CWL Technical Memorandum 23-6, 1958.

In CWL Technical Memorandum 23-6, 1958, it is stated that at the Twelfth Tripartite Conference a U.K. delegate asserted that the U.S. Report CWLR 2148 concerning the effects of V agents on the isolated rabbit heart was in error and that the reported effects were caused by the propylene glycol in which the stock V agents were dissolved before subsequent dilution with saline. Since the chief experimental variable between U.S. and U.K. work was this use of propylene glycol, it was suggested by the U.K. delegate that differences between U.S. and U.K. results might be due to this compound.

In CWLR 2148, control curves showed changes in both heart rate and amplitude of contraction. The rate was reduced to 88% and the amplitude to 70% of normal in 30 minutes. Comparable U.K. controls gave a mean reduction in rate to 92% of normal in 75 minutes with no significant change in amplitude.

In Technical Memorandum 23-6, the U.S. have repeated the control experiments to determine the effect of propylene glycol on the heart, and now find a slight increase in the amplitude of contraction. These results thus differ from those reported in CWLR 2148 and are comparable with U.K. findings. Also VX (0.05/ug/ml) is now shown to reduce the amplitude of contraction by a mean of 19% (3 hearts) as compared with a mean of 5% (5 hearts) which was reported in CWLR 2148.

REFERENCES

1. De Candole, C.A., Douglas, W.W., Evans, C.L., Holmes, R., Spencer, K.E.V., Torrance, R.W., and Wilson, K.M. (1953). Brit. J. Pharmacol., 8, 466.

2. Daly, M. de Burgh, and Wright, P.G. (1956). J. Physiol., 133, 475.

3. Quilliam, J.P., and Strong, F.G. (1949). Brit. J. Pharmacol. 4, 168.

4. Salerno, P.R., and Coon, J.M. (1949). J. Pharmacol., 25, 240.

5. Wilson, K.M. (1949). P.T 152.

6. Briscoe, S., and Burn, J.n. (1954). Brit. J. Pharmacol., 9, 42.

7. Larson, E., and Brown, R.V. (1957). C.W.L.R. 2148.

8. Barnes, J.M., and Duff, J.I. (1954). Brit. J. Pharmacol., 9, 153.

9. Stewart, W.C. (1952). Brit. J. Pharmacol., 7, 270.

10. McEwen, L.M. (1956). J. Physiol., 131, 678.

11. Burn, J.H., and Rand, M.J. (1958). B.M.J. (1), 137.

12. Bertler, A., Carlsson, ., and Rosengren, E. (1956). Naturwissenschaften, 43, 521.

13. Briscoe, S., and Burn, J.H. (1954). J. Physiol., 126, 181.

14. Hoffmann, F., Hoffmann, E.J., Middleton, S., and Talesnik, J. (1945). Am. J. Physiol., 144, 189.

15. Bulbring, E., and Burn, J.H. (1949). J. Physiol., 108, 508.

16. Burn, J.H., and Kottegoda, S.R. (1953). J. Physiol., 121, 360.

17. Hutter, O.F., and Trautwein, W. (1956). J. Gen. Physiol., 39, 715.

18. Marshall, J.M., and Vaughan Williams, E.M. (1956). J. Physiol., 131, 186.

19. Burn, J.H., and Rand, M.J. (1958). J. Physiol., 142, 173.

20. Burgen, A.S.V., and Terroux, K.G. (1953). J. Physiol., 120, 449.

21. Bulbring, E., Kottegoda, S.R., and Shelley, H. (1954). J. Physiol., 123, 204.

22. Creasey, N.H. P.T.P. 650.

23. Martha, E.F., and Wills, J.H. (1955). M.L.R.R. 361.

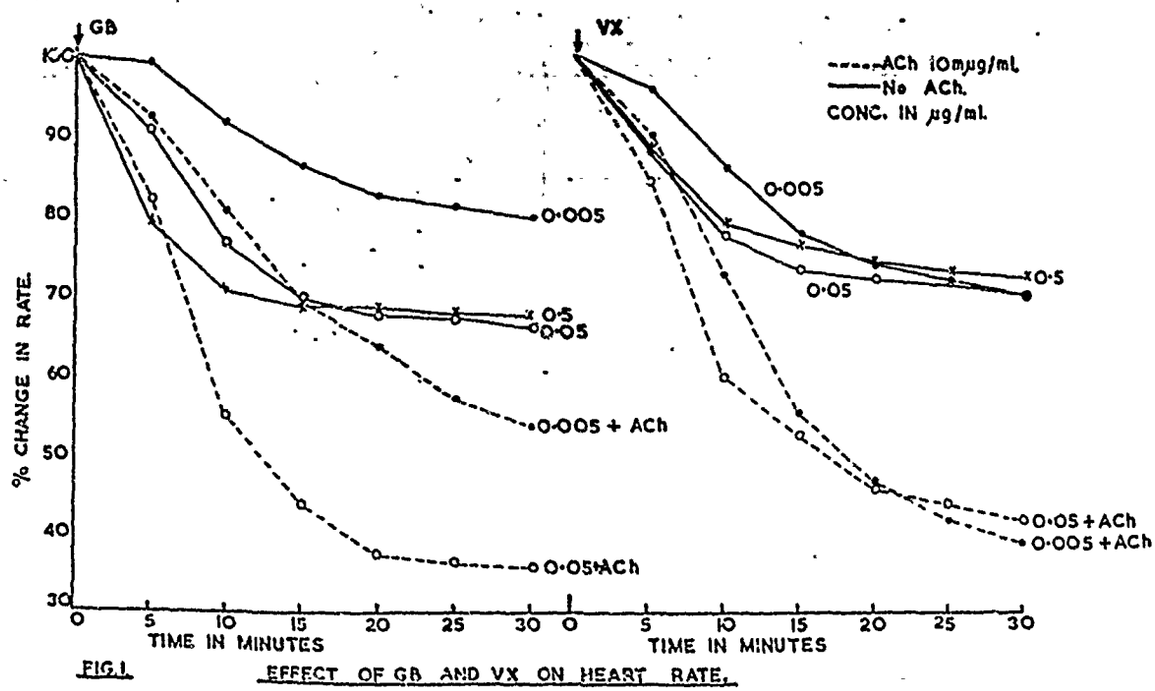

FIG 1 EFFECT OF GB AND VX ON HEART RATE.

PORTON TECHNICAL PAPER 643.

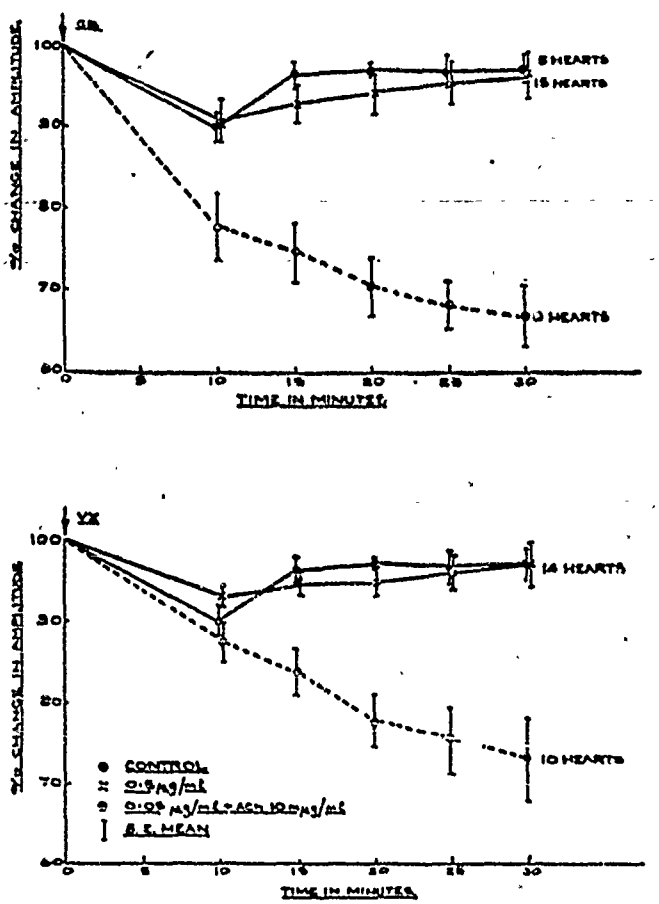

FIG 8 EFFECT OF GB AND VX ON AMPLITUDE OF CONTRACTION.

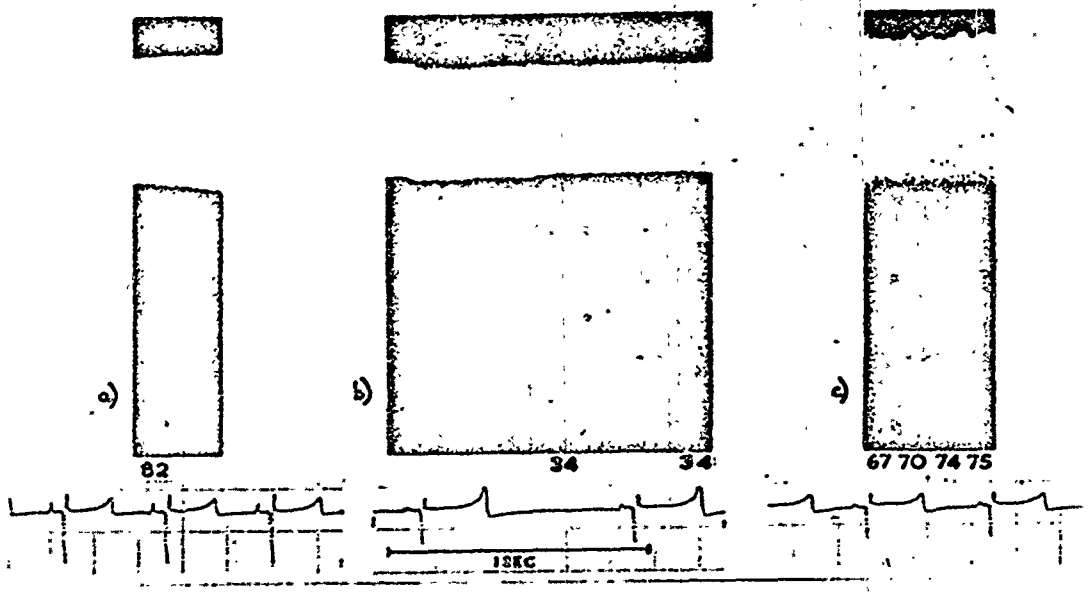

Fig. 1. Perfusion of heart with VX 0.06 /ug/ml + ACh 10 m/ug/ml, followed after 30 min by PZ3 55 /ug/ml + ACh 10 m/ug/ml. Tracings from above downwards: heart contractions, coronary flow, time = 30 sec, heart rate/30 sec, ECG. a) Control; b) 20 - 25 min after commencement of perfusion with VX + ACh; c) 10 min after b), 8 min after changing to perfusion with PZ3.

PTP 643

T1479/1

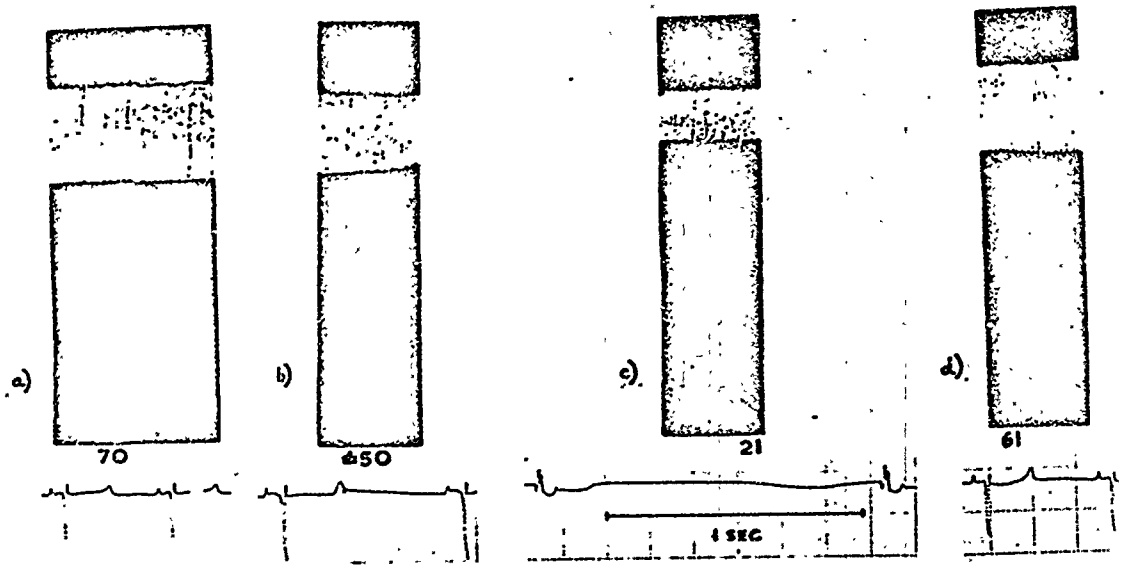

Fig. 4. Perfusion of heart with GB 0.06 /u-e/ml + ACh 10 m/ur/ml followed after 30 min by atropine 2 /e/ml + ACh 20 m/ug/ml. Tracings from above downwards; heart contractions, coronary flow, time = 30 sec, heart rate/30 sec, ECG; a) Control; b) 5 min after commencing perfusion with GB + ACh; c) 10 min after b); d) 20 min after c), 6 min after changing to perfusion with atropine.

PTP 643

T1479/2

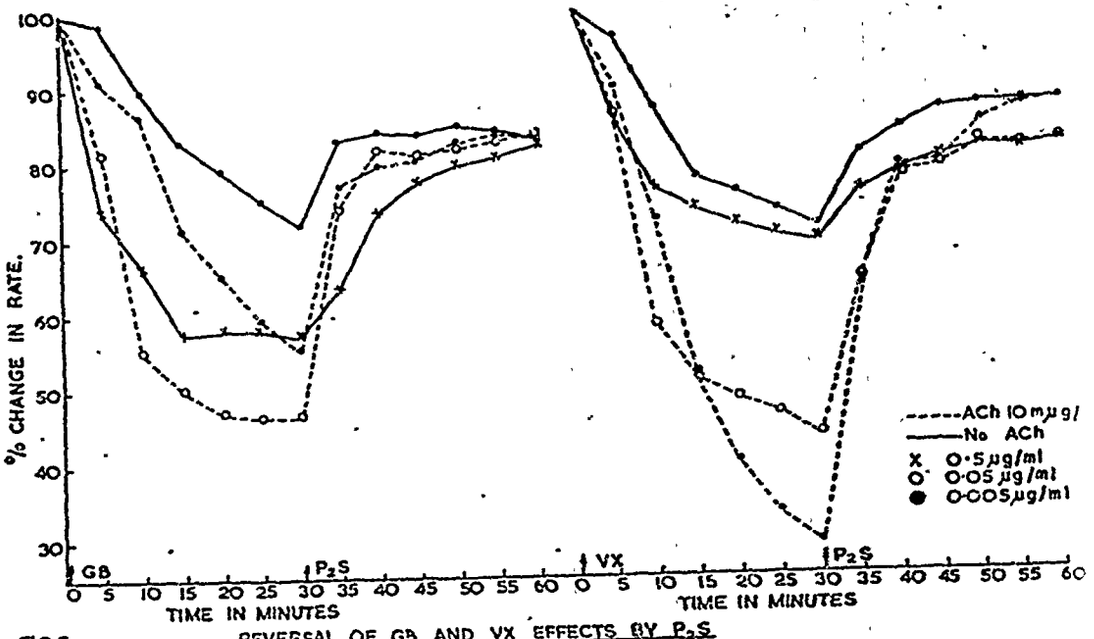

FIG 5 REVERSAL OF GB AND VX EFFECTS BY P_2S

SECRET

DISTRIBUTION

P.T.P. No. 643

MINISTRY OF SUPPLY

Headquarters

1 D.G.S.R.(M)
2-6 D.C.D.P.D.
7 D.P.B.R.
8 Pats.1
9-10 T.I.L.
11-12 File

R. & D. Establishments

90-117 C.D.E.E.
13-14 M.O.S. Estab. Nancekuke
15 C.S.E.E.

CHEMICAL DEFENCE ADVISORY BOARD

16 Prof. H.J. Emeléus
17 Prof. N.K. Adam
18 Prof. D.H.R. Barton
19 A.E. Childs, Esq.
20 Prof. Sir Howard Florey
21 Prof. J.H. Gaddum
22 R.S. Odd, Esq.
23 Prof. Sir Rudolph Peters
24 Sir Eric Rideal
25 Prof. R.H.S. Thompson
26 Prof. D.D. Woods

BIOLOGY COMMITTEE

27 Dr. G.S. Davis
28 Prof. P.R. Allison
29 Prof. E. Boyland
30 Prof. F. Dickens
31 Prof. W.D.M. Paton
32 Sir Bentley Purchase
33 Prof. Max Rosenheim
34 Prof. Andrew Wilson
35-49 Secretariat S.A.C.

BRITISH JOINT SERVICES MISSION

50-56 W.N. Hewson, Esq., M.O.S. Staff

WAR OFFICE

57-58 D.W.D. (G.S.(W)7)

OVERSEAS (through T.I.L)

AUSTRALIA

59-61 Defence Research Laboratories
62 Senior Representative, Dept. of Supply
63 Army Branch Representative
64 R.A.A.F. (Tech. Section)

CANADA

65-66 Chairman, Defence Research Board
67-68 Defence Research Laboratories, Ottawa.
69 Suffield Experimental Station

U.S.A.

70-82 Reading Panel
83-89 U.S. Chem. Corps. Liaison Officer, Porton

SECRET

[dstl]

[dstl]

Defense Technical Information Center (DTIC)
8725 John J. Kingman Road, Suit 0944
Fort Belvoir, VA 22060-6218
U.S.A.

AD#: AD0302603

Date of Search: 8 Oct 2009

Record Summary: WO/195/14431
Title: Biology Committee: The Effects of GB and VX on the Isolated Rabbit Heart and Their Reversal by P2S and atropine
Availability Open Document, Open Description, Normal Closure before FOI Act: 30 years
Former reference (Department): AC 14440
Held by: The National Archives, Kew

This document is now available at the National Archives, Kew, Surrey, United Kingdom.

DTIC has checked the National Archives Catalogue website (http://www.nationalarchives.gov.uk) and found the document is available and releasable to the public.

Access to UK public records is governed by statute, namely the Public Records Act, 1958, and the Public Records Act, 1967.
The document has been released under the 30 year rule.
(The vast majority of records selected for permanent preservation are made available to the public when they are 30 years old. This is commonly referred to as the 30 year rule and was established by the Public Records Act of 1967).

This document may be treated as UNLIMITED.